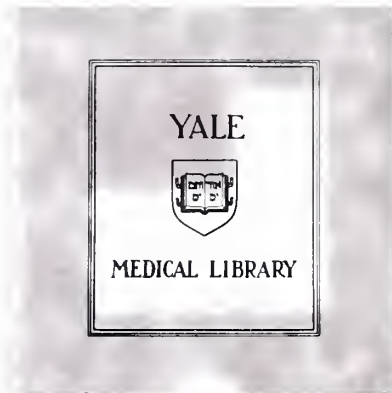HISTAMINE RECEPTORS IN THE
AIRWAYS OF HEALTHY HUMAN SUBJECTS

STEVEN MARK BROWN

1981

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Histamine Receptors in the Airways of Healthy Human Subjects "

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HISTAMINE RECEPTORS IN THE
AIRWAYS OF HEALTHY HUMAN SUBJECTS

By Steven M. Brown

A Thesis Submitted to the
Yale University School of Medicine
in Partial Fulfillment of the
Requirement of the Degree of
Doctor of Medicine

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TO MY PARENTS

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INTRODUCTION

Several chemical mediators play an important role in the normal functioning of the respiratory system in man, in addition to the expression of the immune response. These mediators include histamine, prostaglandins, kallikrein, eosinophil chemotactic factor of anaphylaxis (ECF-A), slow-reacting factor of anaphylaxis (SRS-A), and platelet-activating factor (PAF). All may be released by normal respiratory tissue after passive sensitization with IgE antibody and challenge with specific antigen (8).

Mediators may be released from cells such as mast cells and basophils in response to a number of immunologic or non-specofic stimuli. Airway-constricting stimuli include histamine, cholinergic agonists, SRS-A, bradykinin, prostaglandin $F_{2\alpha}$, β-adrenergic blocking drugs, cold air, dust, sulfur dioxide, exercise, and suggestion (15). In addition to their role in normal physiologic processes, mediators play an important role in the pathogenesis of disease states such as asthma and the anaphylactic response.

The following pages will focus on histamine, a

well-investigated mediator. The actions and role of histamine in both the normal and pathologic setting will be addressed. There will be particular focus on the presence of histamine in the respiratory system. The final chapter includes an original investigation of the presence of histamine receptors in the airways of healthy, human subjects.

HISTAMINE

Background and History

Histamine, β-imidazolylethylamine, was first synthesized in the laboratory in 1907. In vivo, it is formed by the decarboxylation of L-histidine, by histidine decarboxylase. Histamine was first isolated in bacteria in 1910 and was one of the first vaso-active substances to be identified (42, 43). The name is derived from the Greek word histos, meaning tissue.

Histamine is stored in mast cells in various tissues as well as in basophils in blood. The mast cell, which also contains heparin, is the major repository for histamine. Histamine is also present in as yet unidentified non-mast cells of the gastric and intestinal mucosa and the brain. In the rabbit (12), the bulk of histamine in the blood is stored in the platelets.

Histamine is released in response to various immunological stimuli as in IgE-antigen-mediated immune reactions (14). It is also released in response to nonimmunological stimuli such as trauma, toxins, or suggestion (131). The liberation of histamine by cells in response to injurious stimuli was noted as early as 1927 by Lewis (52). Mast cell liberators, such as dextran and anaphylatoxin, may also release histamine.

During the release of histamine from mast cells, the mast cell degranulates. The histamine stored in the granule, along with other preformed and stored mediators, is expulsed from the cell and is followed by release of histamine from a heparin-protein complex (12). A highly polar molecule at physiologic pH, histamine does not diffuse across the blood-brain barrier or cell membranes.

The action of histamine is rapid. It quickly diffuses into tissues and is rapidly metabolized. Gut flora in man converts ingested histamine to the inactive N-acetylhistamine (52). Once released, histamine will disappear from the blood stream within minutes.

Histamine and Its Receptors

Histamine exerts its effects through interaction with at least two identifiable receptors, which are referred to as H_1 and H_2 receptors. These receptors

may be preferentially blocked by specific antihistamines.
The H_1 class of antihistamines are referred to as the
"classical antihistamines". The first antihistamines
to be developed, Bovet and Staub discovered the first
H_1 antihistamines in 1937. The structure is of the form:

$$\begin{array}{c} Ar_1 \\ \\ Ar_2 \end{array} \!\!\!\! \diagdown\!\!\!\!\diagup \; X-C-C-N \diagup\!\!\!\!\diagdown \begin{array}{c} CH_3 \\ \\ CH_3 \end{array}$$
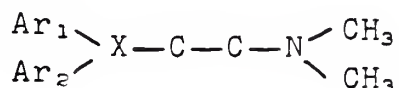
where X may be C, O, or N, Ar_1 and Ar_2 are aromatic or
aryl ring structures and N must be charged at physio-
logic pH. Antihistamines act by competitive and re-
versible occupation of histamine receptor sites without
themselves initiating a response.

Ash and Schild (6) were the first to demonstrate
the presence of more than one class of histamine recep-
tor. They demonstrated that several effects of hista-
mine, such as stimulation of gastric secretion and
inhibition of rat uterus contractions could not be
suppressed by classical antihistamines.

Black, et al. (18) were the first to develop
burimamide, an H_2 receptor antagonist. It was noted
that while burimamide did not have any significant H_1
receptor antagonism, it successfully prevented gastric
acid secretion.

Further evidence for the existence of two separate

classes of histamine receptors has come from the synthesis of agonists specific to H_1 and H_2 receptors. For example, while 2-methylhistamine can elicit the H_1 effect of stimulating bronchial contraction, it has no significant effect on gastric acid secretion. On the other hand, 4-methylhistamine can increase gastric acid secretion, but has little effect on airway constriction (52). In one study (18), 2-methylhistamine was shown to have 16.5% of the H_1 activity of histamine as measured by ability to constrict guinea pig terminal ileum. Its H_2-mediated ability to stimulate isolated atrium was only 4.4%. While 4-methyhistamine had 43% of the ability of histamine to stimulate the guinea pig isolated atria, the relative activity on terminal ileum was only 0.23%.

Other H_1 agonists include 2-(2-aminoethyl)-pyridine and 2-(2-aminoethyl)-thiazole (125). Other H_2 agonists are clonidine (7), St 600 ([2-(5-flouro-o-toluidine)-2 imidazoline hydrochloride])(27), and dimaprit ([S-[3-(N,N-dimethyamino)propyl]isothiourea])(121).

THE ACTIONS AND ROLE OF HISTAMINE AND
ANTIHISTAMINES IN PHYSIOLOGIC PROCESSES

Histamine was recognized a half-century ago (11) as being involved in the inflammatory response and allergic phenomena. The "classic" histamine response

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in man includes itching, swelling, redness (histamine flush), headache, and bronchoconstriction. Histamine will elicit the cutaneous "triple response" of a red spot and flush, wheal, and itch--symptoms which are associated with the inflammatory response and anaphylaxis. The following are brief outlines of some other actions of histamine, with emphasis, where possible, of the pharmacologic effects of histamine and antihistamines in man. The role of histamine in the immune response and pathologic processes will be dealt with in greater detail in a subsequent chapter.

Respiratory System

In man, large concentrations of histamine have the primary effect of bronchoconstriction. Histamine causes an increase in respiratory air resistance and a decrease in airway compliance (8). Other animal species may exhibit either bronchoconstriction, bronchodilation, or both in response to histamine. The response will be determined by which part of the respiratory system is stimulated, which specific receptors are agonized, and what are the effects of those specific receptors. This will be examined in a forthcoming section.

Circulatory System

As in the respiratory system, vasodilation or vasoconstriction will be the effect of histamine agonism,

and will vary depending upon the size and location of the blood vessel, the classification of histamine receptor which is being agonized, as well as the species involved. In man, histamine will cause constriction of large arteries and veins and dilation of the smallest vessels to contain smooth muscle. Intravenous histamine in man, cats, dogs, and sheep will cause a fall in systemic blood pressure secondary to peripheral vasodilation (1, 19).

In cats, dogs, and chickens, the blood vessels dilate in response to both H_1 and H_2 agonism (19, 34, 145). In rabbit blood vessels, H_1 agonism causes vasoconstriction and H_2 agonism causes vasodilation (34). The reverse is the case for calves (34, 66).

In man, peripheral vasodilation is mediated by both H_1 and H_2 receptors (14, 99). Combinations of H_1 and H_2 antagonists are more successful in preventing the effects of histamine on skin (101) and the cardio-vascular system (13), than use of any one antagonist alone.

Vasodilation in the cerebral blood vessels in man accounts for histamine-induced headache.

Histamine's evanescent effect on small blood vessel dilation leads to enhancement of the body's microcirculation, which is due, in part, to the con-

tinuous alternating vasodilation and vasoconstriction caused by changing concentrations of histamine. Histamine will increase capillary permeability with the resultant effect of formation of edema, increased blood viscosity, and hemoconcentration.

The effect on the heart is to stimulate cardiac contractility. Histamine will also increase the rate of cardiac contraction, and will decrease AV-node conduction. In vitro studies of the guinea pig heart (92) show that H_1 receptors may be involved in mediation of a negative dromotropic effect, an atrial inotropic effect, and promotion of histamine-induced arrythmias of conduction. H_2 receptors mediate positive chronotropic effects, ventricular inotropic effects, and promote histamine-induced arrythmias of automaticity.

Histamine will constrict smooth muscle of the spleen.

Endocrine system

The adrenal medulla will release catecholamines in response to histamine.

Tissue Growth and Repair

Histamine is found in increased concentration in areas of wound repair. Increased levels of histamine may be found in regenerating liver, granulation tissue, and the fetus. Tissue damage through local irritants

will increase histidine decarboxylase activity at the
site on injury, leading to increased histamine levels
(12). Histamine promotes wound healing as well as the
formation of keloids and scar tissue. It also helps
to provide an increased blood supply to areas of injury.

Gastrointestinal system

Acting through stimulation of H_2 receptors,
histamine stimulates gastric acid secretion by the
parietal cells of the stomach. H_2 receptor blockade,
in addition to blocking gastric acid secretion, also
blocks the effects of gastrin and pentagastrin. This
finding suggests that histamine may act as a final
"common messenger" for several hormones in the gastric
mucosa (34). Histamine may also stimulate, via H_2
receptors, pancreatic secretion and cholecystokinesis
(34). It will also contract smooth muscle of the gut
via H_1 receptor stimulation.

Nervous system

The function of histamine in the brain remains
obscure, although its presence has been demonstrated
in a variety of structures. The binding of radio-
active H_2 antagonists has been demonstrated in guinea
pig midbrain, cortex, hippocampus, thalamus, brainstem,
hypothalamus, cerebellum, and corpus striatum (23).
Histamine has been isolated in rat, mouse, and monkey

hypothalamus as well as midbrain, cortex, and cerebellum (13). Histamine agonism with the H_2 receptor increases levels of cyclic AMP in these structures (14).

Histamine stimulates sensory nerve endings, producing pruritis. Histamine may facilitate ganglionic transmission via H_1 agonism. H_2 receptors may inhibit ganglionic transmission.

Urinary system

Histamine has a dual mode of action in the human kidney. It causes a pressor response on renal vasculature via H_1 agonism and a depressor response via H_2 agonism.

H_1 Antihistamines

H_1 antihistamines, which include chlorpheniramine, diphenhydramine, and mepyramine, are of therapeutic use in seasonal rhinitis, allergic dermatoses, itching pruritides, hay fever, insect bites, ivy poisoning, blood transfusion reactions, serum sickness, allergic reactions to drugs, Ménière's disease, and vertigo (13, 52). They inhibit the histamine-induced increase in capillary permeability during anaphylaxis. H_1 antihistamines do not completely block anaphylaxis in man because of the significant contribution of several other mediators.

H_1 antihistamines may also cause somnolence,

prevent motion sickness, provide a mild anticholinergic
effect, and have a mild anesthetic effect. Because of
their mild anticholinergic, antiadrenergic, and anti-
serotonergic activities, the use of H_1 antihistamines
in laboratory investigations has occasionally resulted
in erroneously ascribing cholinergic, adrenergic, or
serotonergic effects to histamine.

H_1 antihistamines are metabolized by the liver and
degradation products are almost completely excreted
within 24 hours. Tissues are almost totally free of
H_1 antagonists within 6 hours.

H_2 Antihistamines

H_2 antihistamines have gained recent wide clinical
use in the treatment of a variety of gastrointestinal
disorders associated with hypersecretion of gastric
acid. Of particular significance is the recent popu-
larity of cimetidine in the treatment of duodenal
ulcers.

Another class of histamine antagonist is the mast
cell stabilizer, such as cromolyn sodium. Cromolyn,
which can also inhibit antigen-induced production of hist-
amine (52), has as its main effect, the prevention of
release of histamine and SRS-A. Cromolyn sodium does
not interact directly with either H_1 or H_2 receptors.

THE PRESENCE AND ROLE OF H_1 AND H_2 RECEPTORS IN THE RESPIRATORY SYSTEM

The laboratory investigation which will be described in a forthcoming chapter will examine the presence of histamine H_1 and H_2 receptors in the airways of healthy human subjects. The following is a detailed description of studies which have investigated the presence of histamine receptors in the respiratory systems of a variety of species, both in vivo and in vitro.

Histamine Receptors in Guinea Pig Airways

Guinea pig trachea may be caused to contract due to antigen-induced anaphylaxis (Schultz-Dale phenomenon). It was noted almost fifty years ago (11) that these contractions are associated with the release of histamine from guinea pig lung. The causative effect of histamine in producing the contractions has been demonstrated by the partial or complete block of these contractions by H_1 antihistamines (38, 85). The fact that mepyramine only partially prevents antigen-induced contractions indicates the presence of other mediators which cause tracheal contractility.

Direct stimulation of guinea pig tracheobronchial tissue by histamine results in a profound net smooth muscle contraction (52). Mepyramine totally blocks histamine-induced contraction (85). Metiamide, which

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antagonizes H_2 receptors, potentiates histamine-induced contraction. However, stimulation of H_1 receptors by the specific H_1 agonist, 2-methylhistamine, causes constriction which is not potentiated by metiamide (117). It can thus be inferred that histamine causes both H_1 stimulated contraction and H_2 stimulated relaxation of guinea pig tracheobronchial muscle. Each effect can be isolated by use of specific antagonists.

The H_1 constricting effects and H_2 relaxing effects on guinea pig tracheobronchial muscle can be demonstrated indirectly through studies with antihistamines. However, more direct evidence is available through observation of the effects of histamine agonists. As previously mentioned, the effect of 2-methylhistamine is to cause tracheobronchial contraction.

Another H_1 agonist is 2-(2-pyridyl)-ethylamine (2-PEA). 2-PEA will cause contraction of guinea pig tracheal spirals, in vitro. This constriction of tracheal spirals and parenchymal strips is reversed by dimaprit, an H_2 agonist, which mediates relaxation of the smooth muscle. The effect of dimaprit may be blocked by metiamide (54).

In guinea pig bronchial smooth muscle undergoing ovalbumin antigen-induced anaphylactic contractions, dimaprit had a significant bronchodilatory effect (32).

This effect was even more profound than treatment with an H_1 antagonist, mepyramine.

Direct evidence for histamine H_1 receptor presence in guinea pig lung and other organ systems has come from studies which trace histamine receptors by radioactively labelling tissues with $[^3H]$-mepyramine (41).

Drazan and Schneider (53) found that histamine induced constriction in both guinea pig trachea and lung parenchyma. They discovered that histamine H_1 antagonism by mepyramine was more successful in blocking constriction in lung parenchyma than in trachea. This suggests that histamine receptors are not necessarily distributed evenly in the airways. There appears to be a greater number of receptors and/or sensitivity to the effects of histamine in peripheral airways in the guinea pig. A future study involving radioactively-labelled antagonists in different parts of the respiratory system would be useful in supporting these findings.

In addition to the effect of the direct interaction of histamine with its receptors, histamine receptor agonism may also cause release of prostaglandins. These prostaglandins act as additional mediators of airway reactivity, but do not act as the final mediators of histamine agonism. The mechanism for release of prostaglandins in response to histamine stimulation involves H_1 receptor-cyclic GMP stimulated release of $PGF_{2\alpha}$ and H_2 receptor-cyclic AMP stimulated release of PGE.

Yen, et al. (156) used the presence of prostaglandins

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$PGF_{2\alpha}$ and PGE as an assay for determining the distribution of histamine H_1 and H_2 receptors within the lung. They found that the H_1 antagonist, pyrilamine, blocked the histamine-induced increases in $PGF_{2\alpha}$. Metiamide diminished the histamine-induced release of PGE.

The authors found that in response to histamine in guinea pig trachea, PGE was present in much greater concentrations than $PGF_{2\alpha}$. In guinea pig peripheral airways, $PGF_{2\alpha}$ was present in higher concentrations than PGE (156). The authors conclude that there might be more H_2 receptors in central than peripheral airways. They find this conclusion consistent with the physiologic importance of maintaining the patency of central airways. The rapid shifts, on the other hand, of peripheral airway size through bronchoconstriction, has been suggested to maximize ventilation-perfusion ratios (156).

In additional to prostaglandins, levels of cyclic nucleotides are a useful assay for determining the presence of histamine receptors. In guinea pig lung anaphylaxis, the antigen-antibody reaction yields an increase in levels of cyclic AMP, cyclic GMP, and the ratio of cAMP to cGMP. Pyrilamine inhibits histamine stimulated increases in cyclic GMP (103). Burimamide had no effect on cGMP but blocked the histamine-induced increases in cAMP.

While there may exist different distributions of H_1 and H_2 receptors within guinea pig airways, H_1 and

H_2 receptors may also differ in sensitivity to histamine. Yen (155) showed that in guinea pig peripheral airway smooth muscles, concentrations of $10^{-10} - 10^{-7}$ M of histamine caused bronchorelaxation which was blocked by metiamide. H_1 receptors had the overwhelming response in concentrations of greater than 10^{-6} M of histamine with the resultant contraction blocked by chlorpheniramine. It appears that H_2 receptors are more sensitive to histamine than H_1 receptors, but in high concentrations, H_1 receptors win out, probably because of greater numbers.

The opposite sensitivities to histamine were observed by Martin and Fertel (102) in guinea pig tracheal rings. They noted that at concentrations of 5×10^{-5} M, guinea pig tracheal rings contracted. However, at concentrations of 10^{-4} M of histamine, the tissue relaxed. Consistent with these observations was the increase in cyclic GMP levels at low histamine concentrations. Increased cyclic AMP levels were noted at high concentrations of histamine. Thus, in trachea, there may be greater sensitivity for H_1 receptors, but greater numbers of H_2 receptors.

In addition to the presence of histamine receptors in guinea pig airway smooth muscle, the presence of histamine H_1 receptors in guinea pig alveolar macrophages

has been demonstrated by both agonist and antagonist studies (50).

Histamine Receptors in the Respiratory System of the Rat and Ferret: Possible Presence of a Subclass of Receptors

Chand and Eyre (34, 40) have done extensive studies on the presence and action of histamine receptors in a number of species. They determined that ferret trachea and bronchus both constricted in response to histamine in vitro. This contraction of smooth muscle was blocked by H_1 antagonism with mepyramine. Rat trachea did not contract in the presence of histamine (40).

Both rat and ferret trachea and bronchi could be made to contract in vitro by exposure to carbachol. When subsequently exposed to histamine in the presence of H_1 blockade, the muscles relaxed. This indicates that an H_2 receptor must have been mediating the tracheo-bronchorelaxation, since the H_1 receptors were blocked. Further evidence comes from studies of direct H_2-agonism by 4-methylhistamine which also caused relaxation of smooth muscle (40).

It is very interesting to note that the relaxation was not blocked by metiamide, burimamide, or cimetidine. This important finding suggests the presence of a sub-class of histamine receptors which are not blocked by conventional H_2 antagonists, yet are stimulated by

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4-methylhistamine and produce relaxation of airway
smooth muscle. The proposed name for this subclass
of receptors is H_{2B} or H_3 receptors.

More Evidence for Histamine$_3$ Receptors: The Airways of the Rabbit

Using a similar methodology for in vitro studies
as Chand and Eyre (40), Fleisch and Calkins (70)
ascertained that histamine induces contraction of
rabbit bronchus. This effect was blocked by H_1
antagonism with pyrilamine. Interestingly, under
similar experimental conditions, partially contracted
rabbit trachea experienced relaxation in the presence
of histamine. This effect could not be prevented by
pyrilamine, burimamide, metiamide, indomethacin, or
propranalol. Thus, histamine-induced tracheal relaxa-
tion does not act through H_1, H_2, prostaglandin release,
or β-adrenergic mechanisms (65, 70).

Chand and Eyre (36) also found that partially-
contracted rabbit bronchus relaxed in the presence of
H_1 blockade. This response was not eliminated by
metiamide. It is thus proposed that rabbit trachea
(70) and rabbit bronchus (36) may contain H_3 receptors
which mediate smooth muscle relaxation.

It is important to note that, as in the guinea pig,
the airways of the rabbit experienced different effects

of histamine stimulation, depending upon whether central or peripheral airways were being agonized. Thus, two closely associated tissues in the same system can be pharmacologically distinct. It is important, therefore, to state specifically which tissues in an airway are being stimulated, when analyzing the results of a histamine study or when designing an investigation.

Histamine Receptors in the Horse: A
Possible Animal Model of Human Asthma

Using their model of carbachol-induced, partially-constricted airways, in vitro, Chand and Eyre (37, 39) determined that histamine or 2-methylhistamine causes tracheobronchial constriction in the horse. Mepyramine prevents this histamine-induced contraction. H_2 agonists such as 4-methylhistamine and dimaprit caused tracheobronchial relaxation in this experimental model. Further support of H_2 stimulation of muscle relaxation comes from treatment of partially-constricted bronchus in the presence of H_1 blockade. This results in relaxation, which could only be the result of H_2 receptor agonism.

The relaxation in horse bronchus was blocked by metiamide and burimamide. However, in horse trachea, metiamide, cimetidine, and burimamide had no inhibitory effect. Perhaps horse trachea also has H_3 receptors.

It has been suggested (39) that the horse may be

a good model for human asthma. Horses suffer from "broken wind", an obstructive respiratory syndrome, also known as equine pulmonary emphysema, which, clinically resembles asthma. Thus, the clinical use of H_2 agonists in asthma may be worth future investigation.

H_1 blockade is only minimally useful in inhibiting anaphylaxis in the sensitized horse (63). As in human anaphylaxis, histamine is only one of several mediators which are involved in the anaphylactic response in the horse. Of importance is the fact that H_2 antagonists such as metiamide and burimamide potentiate anaphylaxis in the horse (39). H_2 antagonists have also been noted to exacerbate anaphylaxis in adult domestic fowl and the calf (39). The mechanism behind this potentiation and its implications in the clinical setting will be dealt with in a forthcoming chapter.

Histamine Receptors in Sheep Airways

Using burimamide, Eyre was the first to discover (14, 61, 62, 64) the presence of H_2 receptors in isolated smooth muscle of sheep bronchi. He found that histamine causes contraction of trachea and the major bronchi in sheep. This effect may be blocked by H_1 antihistamines. In response to histamine, the lesser

bronchi and bronchioles would relax. This effect could
be blocked by burimamide (14, 64). Intravenous histamine
results in a net bronchoconstriction in sheep. The net
effect of inhaled histamine is either bronchodilation
or minimal bronchoconstriction in the sheep (61, 65).

Histamine Receptors in Cat Airways

In in vitro cat trachea partially contracted due
to carbachol, histamine challenge leads to a net broncho-
dilation (14, 62, 64, 100). This effect may be blocked
by a combination of H_1 and H_2 antagonists in addition
to propranalol. This finding suggests, that in the cat
trachea, histamine acts by both direct action with both
receptors as well as by indirect action through local
catecholamine release to produce relaxation.

The cat bronchus also relaxes in response to
histamine as well as 2-methylhistamine and 4-methylhist-
amine (35). This effect is not reversed, however, by
either H_1 or H_2 antagonism, β-blockade, or prostaglandin
inhibition. thereby suggesting an H_3 mechanism.

Other investigators (96, 123) found that in the
cat bronchus, in vitro, histamine challenge caused
smooth muscle contraction, acting via H_1 mechanisms
which could be blocked by mepyramine. Similarly,
antigen-induced contraction could be abolished by
mepyramine in cat bronchial tissue.

Histamine Receptors in the Canine Respiratory System

Histamine causes bronchoconstriction in vivo in the dog. When delivered histamine size is greater than 10 microns, bronchoconstriction is mostly through a reflex vagal mechanism. There is a direct effect of histamine on smooth muscle receptors if particle size is less than 0.5 microns (128).

Dixon, et al. (51) examined the effect of histamine, 4-methylhistamine, cimetidine, and chlorpheniramine on total lung resistance and dynamic lung compliance in anaesthetized dogs. While histamine caused an increase in total lung resistance and a decrease in dynamic lung compliance, the H_2 agonist 4-methylhistamine had no such effect. Accordingly, the H_1 antagonist, chlorpheniramine blocked the effect of histamine, while cimetidine had no protective effect. The authors conclude that histamine acts mostly through H_1 mechanisms in the dog and causes airway constriction. Since dynamic lung compliance and total lung resistance are particularly useful in measuring large airways pulmonary function (98), perhaps this conclusion should not be extended to include the peripheral airways of canines. The counterargument is that at 0.5 microns in diameter, the histamine particles were delivered to the peripheral airways (45). Other investigators (5, 33, 83, 132, 153) have found similar

in vitro and in vivo evidence in peripheral and central airways of dogs. The possibility of regional differences in histamine receptor distribution are similarly supported by scientific evidence (33).

Some investigators (153, 154) have found that cimetidine tended to increase airway resistance, suggesting a small H_2-mediated bronchodilatory effect of histamine in the dog lung. On the other hand, Irvin and Dempsey (82) find a minor contribution of H_2-mediated bronchoconstriction in the peripheral airways of dogs. They concur with other authors regarding the absence of H_2 receptors in canine central airways.

As in humans, mepyramine does not completely block antigen-induced anaphylaxis (33), suggesting the role of other mediators such as SRS-A and serotonin in this response. Whereas metiamide has been shown to exacerbate anaphylaxis in the horse, calf, and domestic fowl (39), the H_2 antagonist has no such in vivo effect in immediate hypersensitivity reactions (33, 153) or in vitro models of canine asthma (5, 33). In ovalbumin-sensitized dogs (5), isolated trachealis muscle contracted in response to histamine. This was inhibited by pyrilamine. H_2 blockade with metiamide had no effect.

Of interest is the effect of H_2 stimulation on respiratory secretions, rather than respiratory muscle

activity. Cimetidine blocked stimulation of canine
secretory activity in the lung in histamine-induced
asthma. Chlorpheniramine also had this effect, but
to a smaller extent (153). The investigators conclude
that histamine acts to promote canine asthma through
an H_1-mediated muscular effect and an H_2-mediated effect
on secretory activity of the respiratory tract (153,
154). They propose that H_2 antihistamines might have
therapeutic value in decreasing respiratory secretions
in the asthmatic.

Histamine Receptors in the Airways of Rhesus Monkeys

One published investigation of histamine receptors
in the airways of rhesus monkeys examined the effect
of H_1 and H_2 antagonism on in vivo pulmonary function
(76). The authors measured peak expiratory flow, total
pulmonary resistance, dynamic lung compliance, tidal
volume, and respiratory frequency.

The results of the study indicated that pretreat-
ment with the H_1 antagonist diphenhydramine inhibited
histamine-induced bronchoconstriction. Metiamide
potentiated histamine's bronchoconstrictory effects
when given prior to histamine challenge (76). Thus,
this study seems to indicate that histamine causes
bronchoconstriction in the rhesus monkey via H_1 agonism.
H_2 receptors are present and apparently modulate the

bronchoconstriction, perhaps via relaxation of smooth muscle or by some feedback mechanism.

Histamine Receptors in the Airways of Man

There have been a few recent studies which have investigated the role of histamine receptors in the airways of man. These preliminary reports include studies of healthy human subjects as well as human asthmatics, who represent a special class of people who are exquisitely sensitive to minute quantities of histamine.

Histamine alone causes a net bronchoconstriction in man. Normal subjects receiving histamine have been reported (110) to experience an increase in respiratory system resistance and closing volume and a decrease in vital capacity (VC), forced expiratory volume in one second (FEV_1), and the ratio of FEV_1 to VC. These parameters suggest both large and small airways constriction due to histamine.

Chlorpheniramine clearly prevents histamine-induced bronchospasm in children (137) and adults (50, 71, 99).

Maconochie, et al. (99) found that in healthy subjects, 8 mg. or oral or 5 or 10 mg. of intravenous chlorpheniramine generally prevented histamine-induced bronchoconstriction, as measured by FEV_1.

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However, 400 mg. or oral cimetidine or 100 or 200

mg. of intravenous cimetidine blockade of H_2 receptors

had no effect on bronchoconstriction in the subjects

studied (99). These findings indicate the primary and

perhaps sole involvement of H_1 receptors in histamine-

induced bronchospasm in healthy, human airways.

The authors offer (99), as an explanation for the

lack of effect of cimetidine, the possibility that

cimetidine did not reach the H_2 receptors in the lung

in sufficient concentrations to produce an effect.

However, the doses of cimetidine used were adequate to

inhibit gastric acid secretion and histamine challenge

did coincide with peak blood levels of cimetidine.

Eiser (59) was similarly not able to demonstrate

the presence of H_2 histamine receptors in normal human

airways. He found that H_2 blockade with 200 mg. of

intravenous cimetidine had no effect on histamine-induced

changes in airway specific conductance in the nine

subjects tested. 20 mg. of intravenous chlorpheniramine,

however, significantly shifted the histamine dose-response

curve to the right. Eiser found no difference between

chlorpheniramine administration and the administration

of chlorpheniramine and cimetidine together (59).

Cimetidine has received wide clinical use since

its development. In the clinical setting, it does not

appear that cimetidine promotes asthmatic bronchospasm (14).

However, Frith, et al. (71) are preparing a report
on a study of ten asthmatics. The subjects were selected
on the basis of histories of episodic dyspnea as well as
documented reversible airflow destruction. Subjects
received an increasing dose of inhaled histamine until
a 20% drop in FEV_1 was reached. On subsequent days,
the same protocol was followed, except subjects were
pretreated with chlorpheniramine 8 mg. or cimetidine
600 mg. or both.

The findings show that H_1 blockade significantly
increased the level of histamine necessary to produce
a 20% drop in FEV_1 (p=0.0001)(71). Cimetidine did not
differ from placebo. However, cimetidine did signi-
ficantly (p=0.04) reduce the effectiveness of chlor-
pheniramine when the two drugs were given together.
These results indicate the presence of histamine H_1 and
H_2 receptors in asthmatic airways, although the H_2
effect does not seem to be very profound. The authors
do not address the influence of drug-drug interactions
in their findings. The dose of cimetidine used was
at least twice that required to reduce food-stimulated
gastric acid secretion by 50%.

Nathan, et al. (109) achieved similar results in
eleven asthmatics. Chlorpheniramine significantly

raised (p less than 0.002) the level of histamine required to cause a 20 percent drop in FEV_1, a 35 percent decline in maximum expiratory flow at 50 percent below vital capacity, or a fifty percent decrease in airway conductance. Cimetidine significantly decreased the amount of histamine necessary to produce these results (p less than 0.02). The authors conclude that H_2 receptors, mediating bronchodilation, exist in asthmatic airways.

Pretreatment with aerosolized diphenhydramine in adult asthmatics significantly blocked the effects of inhaled histamine (28) as measured by the amount of histamine required to produce a 20% drop in FEV_1. This result lends further support to the theory that histamine produces bronchoconstriction by direct action on H_1 receptor sites.

Although oral antihistamines are of little clinical use in asthmatics, clemastine, an H_1 antagonist, when delivered directly to the bronchial tree by inhalation (113), caused bronchodilation in asthmatics. Pulmonary function parameters were FEV_1 and peak expiratory flow rate (PEFR). In addition, the histamine-dose response curve was significantly shifted to the right (112). These findings wree comparable to the results achieved by salbutamol, a β_2 receptor agonist. The results of

this study may be due to complete blockage of H_1 receptors in asthmatics, thereby allowing endogenous histamine to react specifically with the only unblocked receptors, H_2 receptors, thereby mediating relaxation in addition to the obvious elimination of bronchoconstricting H_1 activity. An interesting follow-up study would investigate whether cimetidine reverses the bronchodilatory effects of inhaled clemastine.

Dunlop and Smith (57) as well as Kaliner and Platshon (89) have provided in vitro evidence for the presence of histamine receptors in human airways. Dunlop and Smith (57) showed, in vitro, in sensitized human bronchus, 2-3 mm. in diameter, that in the presence of H_1 receptor blockade, histamine agonism led to bronchorelaxation. This effect could be eliminated by H_2 antagonism with metiamide. Furthermore, in human bronchus caused to contract due to exposure to house dust mite antigen (57), H_1 blockade with mepyramine caused decreased contraction while H_2 blockade caused increased bronchial contraction.

Kaliner and Platshon (89) demonstrated that 2-methyhistamine produced an increase in cyclic GMP, with subsequent enhancement of mediator release (SRS-A). H_2 agonism with dimaprit in the human lung caused increased levels of cyclic AMP with subsequent decreased

release of SRS-A. This finding indicates that H_2 agonism with substances such as dimaprit might be useful in the treatment of anaphylaxis.

Histamine's action in asthmatic airways to cause bronchoconstriction is almost entirely via direct stimulation of smooth muscle receptors (29). In asthmatics receiving a dose of atropine, a parasympatholytic drug, at a dose strong enough to achieve cholinergic blockade, the histamine dose-response curve was slightly shifted to the right (29, 84, 137). Thus, in asthmatics, histamine does not act primarily through cholinergic pathways.

Similarly, the vagal-blocker SCH 1000 did not prevent histamine-induced bronchospasm in asthmatics (150) although it did prevent methacholine-induced bronchospasm. Histamine does have some reflex vagal nerve irritant receptor action, although this is a minor effect (3, 135). In large airways, it is proposed that histamine has more of a reflex vagal effect (8, 47) while in small airways, constriction is due to direct histamine H_1 antagonism.

As is the case in other species, such as the guinea pig and sheep, the distribution and sensitivity of histamine receptors may vary in different parts of the respiratory system such as in central versus peripheral

airways. This possibility makes _in vivo_ studies in man
more difficult than _in vitro_ studies. Methodologies
must deliver histamine to specific parts of the airway.
Pulmonary function tests must be used to measure constric-
tion or dilation in the specific stimulated sections of
airway under investigation. Thus, one would not want
to only measure large airways constriction in a protocol
where histamine is delivered mostly to the respiratory
bronchioles. Since large airways have a reflex vagal
contribution to histamine-induced bronchospasm (47) this
provides another factor which makes analysis of the presence
of histamine receptors _in vivo_ difficult.

Pulmonary Vascular Histamine Receptors

The most profound effect of histamine on airway
mechanics is by its direct interaction with smooth muscle
receptors. However, interaction of histamine with
pulmonary vascular receptors is also very important in
the regulation of respiration. Studies with a variety of
results have been performed in a number of animal species.

In the guinea pig, histamine agonism leads to
pulmonary vasoconstriction. This effect is most pro-
found in the pulmonary veins (34, 115). It is proposed
(64) that histamine-induced pulmonary venospasm may
increase right atrial pressure and thereby contribute
to the formation of pulmonary edema. The pulmonary

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vasoconstriction appears to be mediated by H_1 receptors in the guinea pig. Burimamide potentiates this response. Mepyramine has a depressor action on pulmonary vasculature, indicating the presence of H_2 receptors which mediate pulmonary vasodilation. Some authors feel that pulmonary vascular dilation may increase airway resistance, through mechanical obstruction.

The rat exhibits pulmonary vasoconstriction in response to hypoxia. This effect is blocked by metiamide (10) indicating that H_2 stimulation leads to pulmonary vasoconstriction in the rat. This is the opposite effect to be observed in the cat (79, 145) where H_1 agonism causes pulmonary constriction and H_2 agonism mediates dilation of pulmonary vasculature.

Comparable results are available in the dog (74). Okpako (116) demonstrated that H_2 agonism with 4-methylhistamine had 15 times the vasodepressor potency of 2-methylhistamine. This vasodepression could be abolished by burimamide.

Tucker, et al. arrive at similar results in the dog (145, 146). Measuring pulmonary vascular resistance, they found that H_1 agonism led to an increase in resistance, while H_2 agonism caused a fall in pulmonary vascular resistance. Antagonists had the expected results of reversal of the effects of agonists. The

authors also found (146) that H_2 blockade would also
potentiate pulmonary vasoconstriction during hypoxia.
Okpako (116) concludes that H_2 receptor agonism may
lead to "pooling of blood" in the bronchial mucosa
with resultant mechanical obstruction of the airway.

The effect of histamine on pulmonary vessels seems
to vary with development in dogs (111). One group of
authors found that while metiamide potentiated pulmonary
vasoconstriction in canine pups, this effect disap-
peared after 15 days of life.

In the horse (77), histamine causes pulmonary
vasoconstriction. This appears to be mostly an H_1
effect, although H_2 agonism may also contribute to
vasoconstriction to a lesser degree. In the nonpregnant
ewe (152), histamine also mediates pulmonary artery
constriction which may be blocked equally well by H_1
or H_2 antagonists (benadryl and metiamide).

THE ROLE OF HISTAMINE IN PATHOLOGIC PROCESSES

Histamine, Inflammation, and the Allergic Response

As early as 1910 (42, 43), Dale and Laidlaw
appreciated the similarities between the effects of
histamine and anaphylaxis. These include redness,
swelling, edema, and hypotension. Twenty years later,
Bartosch et al. (11) demonstrated the presence of
histamine in guinea pig lung during the antigen-antibody
reaction.

Sensitized human lung and skin mast cells and leuko-
cytes will release histamine in vitro when challenged
with specific antigens (17). In vivo, sensitized humans
and asthmatics experience increased plasma histamine
levels following allergen-induced bronchoprovocation
(17, 28). The increase in histamine levels correlates
well with the onset of bronchospasm.

The release is initiated by IgE antibody bound to
the cell surface. IgE-induced effects are mediated by
alterations in cyclic nucleotides.

The fact that a second messenger, in this instance,
the cyclic nucleotides cAMP and cGMP, may act as the final
common mediators for the elaboration of the different
physiological responses of different tissues to antigen-
antibody interaction, may have been first proposed in
1919 (43):

> So that it is quite possible that
> the production by histamine, and by
> a whole group of other substances,
> of a complex including contraction
> of plain muscle with relaxation and
> permeability of capillaries, may
> depend on a common type of physical
> change in protoplasm produced by all
> of them, the result of which receives
> different expression in terms of the
> physiology of different tissues.

Mechanisms of Histamine Interaction

In cells undergoing the IgE mediated antigen-antibody reaction (Type I immediate hypersensitivity reaction), histamine, acting on H_1 receptors, participates in an exquisitely fine-tuned mechanism to promote the inflammatory process. In addition, histamine also acts on H_2 receptors to provide a negative feedback on its own actions (34).

Step #1: Histamine Release

Following the interaction of cell surface-bound IgE antibody with specific antigen, there is an influx of extracellular calcium, important for the initiation of release of histamine-containing granules. This is followed by the energy-dependent activation of microtubules and microfilaments which results in the fusion of the perigranular membrane with the cell membrane, followed by exocytosis of granules. Extracellular ions such as sodium then penetrate into the granules with the resultant release of the histamine-

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heparin-protein complex by ion exchange (114).

Further release of histamine is modulated by intra-
cellular levels of cyclic AMP and cyclic GMP, which, as
will be seen, are themselves regulated by histamine.
Platshon and Kaliner (124) found that cGMP and cAMP
levels increased profoundly, simultaneously with the
appearance of mediators in sensitized human bronchus.

H_1 Receptors Promote the Inflammatory Response

Acting on H_1 receptors, histamine's effect on
smooth muscle is to facilitate contraction by in-
creasing calcium entry. This leads to depolarization
and impulse-carrying currents.

The sudden contraction, which is a result of
histamine release, is followed by a brief period of
partial relaxation during the anaphylactic response.
This is followed by a secondary, sustained contraction,
which is resistant to classical antihistamines and
which is attributed to the delayed release of non-hist-
amine mediators such as SRS-A (38).

Cyclic GMP may be implicated in this H_1 mediated
contraction. Increased levels of cyclic GMP are found
following stimulation with histamine (124). This
increase may be blocked by H_1 antihistamines (124).
The rise in cyclic GMP is calcium-dependent (14). The
role of cyclic GMP may be to activate protein kinases

which in turn enhance phosphorylation of proteins which
participate in calcium ion mobilization (14) which in
turn leads to smooth muscle contraction.

Histamine seems to promote its own release through
H_1-mediated positive feedback (14). H_1 receptor anti-
histamines inhibit the IgE induced release of histamine
as well as antigen-induced release of histamine from
sensitized monkey lung (14).

H_1 receptor agonism enhances eosinophil migration.

Histamine causes release of prostaglandins from
the lung. Indomethacin, a prostaglandin synthetase
inhibitor, diminishes allergic bronchospasm by 40%,
indicating a role of prostaglandins in allergic broncho-
constriction, although the prostaglandin synthesis
which accompanies anaphylaxis has been considered
secondary to the action of histamine. Suppression of
prostaglandin synthesis does not prevent hypersensitivity-
associated rises in cAMP and cGMP.

The cyclic GMP dependent release of $PGF_{2\alpha}$ and
thromboxane A_2, which intensify bronchoconstriction,
may be inhibited by H_1 receptor antagonism with pyri-
lamine or mepyramine (64, 125). H_1 agonism with
2-methylhistamine promotes release of $PGF_{2\alpha}$ and throm-
boxane A_2 (16, 124).

Histamine agonism of H_1 receptors (and to a

lesser extent H_2 receptors) also increases vascular
permeability. This facilitates the migration of immuno-
globulins and leukocytes to the site of inflammation.

H_2 Receptors Inhibit the Inflammatory Response:
Potentiation of Anaphylaxis By H_2 Receptor Blockade

Histamine acts through H_2 receptors in a different
manner than H_1 receptors. Stimulation of histamine
H_2 receptors causes a rise in cyclic AMP (124). Blockade
of the H_2 receptor prevents a rise in cyclic AMP (2,
124, 134, 138), except in mice, where H_1 and not H_2
receptor blockade prevents histamine-induced cAMP
accumulation (118).

Cyclic AMP facilitates calcium sequestration and
extracellular transport, with resultant relaxation of
contractile proteins. Elevated levels of cyclic AMP
correlate with bronchial smooth muscle relaxation
and bronchodilation.

Effect on Histamine Release

H_2 receptor agonism and the subsequent rise in
cyclic AMP is also very important in the regulation
of the immune process. Elevated levels of cyclic AMP
inhibit histamine and and SRS-A release from the mast
cell and human leukocytes (21).

In humans allergic to ragweed, histamine is re-

leased upon exposure to ragweed antigen. In leukocytes of these human donors, in vitro, this release of histamine may be inhibited by histamine, isoproteronol, or methylxanthines. Each of these substances acts to increase levels of cyclic AMP (21). Lichtenstein and Gillespie (94) demonstrated that H_2 antagonists could prevent the inhibition of histamine release from blood basophils. H_1 antagonists did not block this response.

Ovalbumin antigen caused dose-related release of histamine from in vitro guinea pig lung. Dulabh and Vickers (56) found that H_2 antagonism with cimetidine or burimamide potentiated this release. This demonstrates the role of H_2 receptors in modulating the immune response-mediated release of histamine.

In addition, metiamide or cimetidine potentiated the antigen-induced bronchoconstriction in guinea pig lung (56). Other investigators have achieved similar results (2, 90).

It appears that metiamide also potentiates the antigen-induced release of histamine in sensitized rhesus monkey lung (30, 31), canine lung, bovine lung, and monkey skin (30) in vivo. However, Platshon and Kaliner (124) were unable to prove that H_2 blockade potentiated histamine release in the human lung in vitro. H_2 antagonism does not potentiate histamine release from passively sensitized rat lung (30, 34). This

suggests that H_2 agonism may not inhibit endogenous histamine release in the rat (30). Holyroyde and Eyre (80) showed that H_2 blockade actually prevented histamine release in sensitized bovine lung. It is interesting to note that H_2 blockade does not, in some experimental models (30, 33, 88) also enhance SRS-A release from sensitized tissues. This is a curious result, since SRS-A release is inhibited by increased levels of cyclic AMP and H_2 agonism increases cyclic AMP levels.

Effect on Histamine Production, Metabolism, Uptake, Storage, and Clearance

Not only does H_2 agonism prevent release of histamine, it also inhibits production of histamine. H_2 antagonists induce histidine decarboxylase to produce histamine (104, 125). Histamine metabolism is also promoted by H_2 receptor agonism and is inhibited by H_2 antihistamines (67, 107, 125). Taylor (144) was unable to show, however, that either the H_1 antihistamine mepyramine, or the H_2 antihistamine burimamide had any effect on histamine methyltransferase, the enzyme which is involved in the methylation of histamine, the predominant mode of inactivation of the mediator. Blockage of the H_2 receptor also inhibits histamine storage in mast cells (14, 107).

H_2 antagonists prevent histamine uptake (34, 68, 107) and delay clearance of histamine from the circulation (34, 133).

Effect on Other Participants in the Immune Response

Increased levels of cyclic AMP, the result of H_2 agonism, also inhibit T-cell induced cytolysis (14, 126). H_2 agonists in high concentrations will inhibit eosinophil migration (14) as well as inhibit lysosomal enzyme release from human polymorphonuclear cells (93, 125). Acting through the H_2 receptor, histamine will inhibit the production of lymphokines.

Just as histamine acts upon H_1 receptors to promote $PGF_{2\alpha}$ release, histamine acts upon H_2 receptors to promote release of PGE, a prostaglandin which has cyclic-AMP-mediated, bronchodilating activity. PGE may in turn inhibit further histamine release by activation of cAMP in mast cells (16). Metiamide can antagonize histamine-induced release of PGE.

In conclusion, histamine released during a hypersensitivity reaction may participate in a number of mechanisms to provide feedback inhibition in order to limit the severity of the reaction.

Histamine, Antihistamines, and the Immune Reaction in the Clinical Setting

Antihistamines as Preventors of the Inflammatory Response

Although histamine is a major mediator of anaphylaxis, antihistamines have not been effective in the complete control of this process. Several reasons for this have been suggested. Other mediators are involved in the anaphylactic response. These include SRS-A, kinins, prostaglandins, and serotonin. SRS-A has been shown to be the mediator responsible for the late, prolonged phase of broncho-constriction in the antigen-challenged human bronchus (125). Although H_2 receptors seem to be most important in inhibition of the inflammatory response, to the effect that they cause increased vascular permeability, they contribute to the inflammatory process. Classical H_1 antihistamines have no effect on the H_2 receptor contribution to this process.

Antihistamines nevertheless do have important clinical relevance in the modification of the inflammatory reaction. Plaut (125) found that cutaneous reactivity to compound 48/80 could be inhibited by combinations of H_1 antihistamines and cimetidine. He proposes that H_1 and H_2 receptor blockade might be helpful for treating some cutaneous manifestations of allergic diseases such as chronic urticaria (125).

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Similarly, Harvey and Schocket (78) found that H_2 antihistamines potentiated the effect of H_1 antihistamines in blocking the cutaneous wheal response, although H_2 antihistamines alone had no effect. It is suggested (78) that H_2 receptors are somewhat involved in the cutaneous histamine response. Goadby and Little (75) found that metiamide protected against anaphylaxis in the guinea pig, a finding which is unusual, considering the purported role of H_2 receptors in modifying the immune response.

Rocklin (129) noted that in sensitized guinea pigs, histamine in concentrations of $10^{-3}M$ reduced the size of a delayed hypersensitivity skin test. This effect could be reversed by an H_2 antagonist. He proposed that histamine suppresses cutaneous delayed hypersensitivity in part, by inhibiting the production and proliferation of migration inhibiting factor (MIF). H_2 antagonists reversed this inhibition of MIF (129). In so doing, H_2 antagonists such as cimetidine have been shown to be of therapeutic value in allowing the expression of cutaneous cell-mediated immunity. For example, cimetidine is useful in the treatment of selected deficiencies in the immunologic response to candida antigen (86).

Antihistamines as Potentiators
of the Inflammatory Response

H_2 antihistamines, by antagozing H_2-mediated inhibition of inflammation, may potentiate anaphylaxis. Dunlop and Smith demonstrated (57) that metiamide potentiates anaphylactic bronchoconstriction in vitro. This was the observation of Drazen, et al. (55) who found that H_2 antihistamines burimamide and metiamide increased the severity of immune-initiated anaphylaxis in the guinea pig. 4-methylhistamine decreased the severity of the reaction (55). Of note is the fact that cimetidine, another class of H_2 antagonist, had no effect.

Wolfe, et al. (151) came to the same conclusion regarding cimetidine. They were unable to demonstrate a prominent modulating role for H_2 receptors in immediate or delayed skin test reactivity. Krell and Chakrin (90) were unable to find that H_2 blockade with metiamide potentiated antigen-induced or histamine-induced changes in dynamic lung compliance or pulmonary resistance in the dog at doses up to 16 times the ED_{50} for inhibition of gastric acid secretion. They conclude that insofar as the sensitized canine might be a model for human asthma, it appears that H_2 antihistamines may not be deleterious to allergic asthmatics (90).

While in one study, cimetidine potentiated the

anaphylactic response in the guinea pig (56), required doses were 100 micromoles per kilogram, a dose between 50 and 100 times that required to inhibit maximal gastric output by 50% in the rat and dog (14). It is unlikely that the results of Dulabh and Vickers (56) can be extended to conclude that a potential effect of therapeutic doses of H_2 receptor antihistamines would be to cause intensified allergic reactions in man.

However, a recent case report in the New England Journal of Medicine (48), described the appearance of a hypersensitivity reaction which coincided with the oral administration of cimetidine. The reaction disappeared after discontinuance of the H_2 antagonist.

This finding was supported by the work of Avella, et al. (9) who found that in patients receiving cimetidine, there was a significant enhancement of the delayed hypersensitivity response to four common antigens as compared to controls. This is most likely due to the antagonism of H_2 receptor mediated stimulation of suppressor T cell function (9).

Expecting that H_2 blockade would remove inhibition of the delayed hypersensitivity reaction, Dale (44) noted that metiamide did not exacerbate the tuberculin reaction in the guinea pig. She concluded that other factors in addition to histamine must regulate the

immune response.

The Role of Histamine in Asthma

Introduction

Asthma may be defined as a disease process characterized by an increased reactivity of the trachea and bronchi to various stimuli (4). This leads to reversible narrowing of airways. Asthma is marked by increased contraction of bronchial smooth muscle and increased secretion of mucous in the bronchial tree. Asthma is manifested by the symptoms and signs of dyspnea, wheezing, cough, prolonged expiration, and responsiveness to bronchodilator drugs.

Asthmatic airways display hyperreactivity to a number of stimuli. These include specific allergens such as house dust, animal dander, pollen, and drugs (114). Other stimuli include suggestion (81, 97, 131, 140), smoke, excercise (136), respiratory infections (4), and emotional reactions (4).

The mechanism for the allergic hyperreactivity in asthmatics is via the Type I immediate hypersensitivity reaction which is mediated by the IgE antibody which reacts with surface bound antigen. Asthmatic broncho-spasm is also mediated via Type III hypersensitivity, a late, gradually appearing immune-complex reaction.

The most important mediators of asthmatic broncho-
spasm are histamine and SRS-A. Other mediators such
as bradykinin and prostaglandins play a less important
role. Bronchoconstriction is mediated by chemical
mediators, as previously discussed, via a rise in intra-
cellular levels of cyclic GMP. This in turn causes a
rise in intracellular Ca^{++} levels with subsequent
activation of contractile proteins.

In addition to chemical mediators, bronchoconstric-
tion is promoted by increased parasympathetic tone.
Cholinergic stimulation similarly causes a rise in
intracellular calcium ion levels (114). Interaction
of bronchoconstricting elements with "irritant recep-
tors" causes reflex bronchoconstriction through vagal
stimulation. Vagally-induced bronchoconstriction can
not be inhibited by classical H_1 antihistamines, although
it can be prevented by treatment with parasympathetic
blocking agents such as atropine or ipratropium bro-
mide (114).

Agonism with β_2 receptors on respiratory smooth
muscle causes bronchorelaxation. Neurotransmitters
which combine with β_2 receptors will relax bronchial
smooth muscle. The mechanism of bronchorelaxation
is via stimulation of cyclic AMP which causes decreased
calcium ion levels. This increase in cyclic AMP

which results from catecholamine stimulation, also decreases the antigen-induced release of histamine in sensitized lung in vitro (106).

Asthmatics may experience disturbances in any of several mechanisms which lead to increased bronchial activity. They may display hypersensitivity to antigens, increased responsiveness to histamine, abnormally active parasympathetic tone, or diminished sympathetic (β_2 receptor) responsiveness (106, 110, 142). The following pages will focus on the role of histamine in asthma.

The Role of Histamine in Asthma

The findings that histamine causes bronchoconstriction and is released in immediate hypersensitivity reactions have led to a strong association of asthmatic bronchospasm with histamine (117). With regard to histamine, asthmatics may experience an increased release of histamine in response to bronchoconstricting stimuli as compared to healthy humans.

For example, in addition to the role of vagal stimulation in exercise-induced bronchospasm, histamine also seems to be involved. Simon, et al. (136) found that among some asthmatics, excercise provoked a fall in pulmonary function (FEV_1). This decline in FEV_1 correlated well with increased levels of histamine in the peripheral venous blood.

Asthmatics may also have an increased sensitivity to the histamine which is released. Indeed. asthmatics develop bronchoconstriction in response to lower concentrations of inhaled histamine than do normal persons (91, 110, 139). This fact is useful in the identification of persons with hyperreactive airways (109). There is some evidence in the mouse model for asthma, that histamine sensitivity may be in part, an inheritable trait (147).

In addition, asthmatics differ from normals in that asthmatics have detectable levels of histamine in their plasma (17) even without antigen challenge. Nonasthmatics do not have any detectable level of endogenous histamine in their plasma.

Asthmatics might suffer from an imbalance in the numbers of different types of histamine receptors which have opposing effects on bronchial smooth muscle. This theory is supported by recent experimental evidence. Busse and Sosman (26) noted that histamine, acting through H_2 receptors, inhibits serum-activated zymosan-induced lysosomal enzyme release from granulocytes. This response parallels a simultaneous increase in levels of cyclic AMP. The authors found (26) that this H_2-mediated inhibition of lysosomal enzyme release is significantly decreased in asthmatics as well as

normal volunteers infected with rhinovirus 16 (24).
This suggests that asthmatics have decreased numbers
or sensitivity of H_2 receptors. Furthermore, viruses
may exacerbate the asthmatic condition through their
effect on H_2 activity.

The decreased responsiveness of H_2 receptors in
asthmatics is important. The antigen-antibody reaction
might initiate a selective increase in cyclic GMP levels
in asthmatics (124). Since the H_2 receptor acts to
control further histamine release from sensitized cells
as well as acts to stimulate histamine metabolism and
uptake, diminished H_2 reactivity would cause increased
levels of endogenous histamine. In asthmatics, who
are sensitive to even minute quantities of histamine,
this increase in histamine levels would lead to further
bronchoconstriction. For the same reason, H_2 blockade
in asthmatics would be expected to be more deleterious
than in non-asthmatics, because the slight increases
in histamine would effect asthmatics, whose H_1 receptors
are much more sensitive to slight increases in the
levels of histamine.

Airway obstruction in asthma is due, in part,
to inflammatory changes in the bronchi. Granulocytes
may be found in the respiratory tree in increased
numbers during bronchial reactivity. Important medi-

ators of this inflammatory response are the lysosomal enzymes. Decreased inhibition by the H_2 receptor of these lysosomal enzymes may contribute to the inflammatory response in asthmatics (26). It is interesting to note that persons with other defects in immune responsiveness, such as those with atopic eczema, may also have diminished H_2 receptor-mediated inhibition of the inflammatory process (25).

Some authors, however, have found no difference between H_2 receptor responsiveness in asthmatics and non-asthmatics. Gillespie et al. (73) found no significant difference between asthmatics and non-asthmatics in the effect of histamine on lymphocyte cAMP levels (an H_2 receptor-mediated process).

The finding that asthmatics have decreased H_2 receptor responsiveness is analagous to the decreased responsiveness of β_2 adrenergic receptors seen in asthmatics as compared to non-asthmatics (106, 110, 142).

HISTAMINE RECEPTORS IN THE AIRWAYS OF
HEALTHY, HUMAN SUBJECTS

Introduction

While many of the previously described studies
have provided excellent evidence for the presence of
histamine H_1 and H_2 receptors in the airways of many
species, there is little evidence for the presence
of H_2 receptors in human airways. Should H_2 receptors
exist in normal or asthmatic airways, and should they
provide a role in bronchodilation, as in animal models,
this finding might have important therapetic implica-
tions. For example, H_2 receptor agonists might be
useful as bronchodilators. Furthermore, caution might
need to be followed in using H_2 receptor antagonists
in persons with compromised respiratory function.

Evidence to date includes _in vitro_ studies on the
presence of H_2 receptors in sensitized human bronchus
(57, 89). Two recent studies in non-asthmatics (59, 99)
have found no evidence for the presence of H_2 receptors
in the airways of non-asthmatics. Two recent studies
(71, 109) provide preliminary evidence that H_1 and H_2
receptors are present in asthmatic airways, and H_2
receptors mediate bronchodilation.

The following experiment describes an investigation
of the effects of histamine H_1 and H_2 receptor blockade

in the airways of healthy, non-asthmatic, human subjects.
The goal of the experiment was to provide evidence for
the presence or absence of H_1 and H_2 receptors in the
small airways of normal man.

Materials and Methods

Subject Selection

Subjects were recruited as paid volunteers. They
gave written, informed cinsent for participation in
the experiment, as approved bv the Yale University
School of Medicine Human Investigations Committee.

Eleven healthy, non-asthmatic subjects (8 males,
3 females) ranged in age from 18 to 35. The mean age
was 26.3 ± 5.1 years. All had no history of asthma
or recent respiratory diseases which could simulate
an asthmatic response to histamine (24, 60). Baseline
pulmonary functions were at least 85% of normal predicted
values for the group as a whole. (TABLE I)

Histamine preparation

Histamine dihydrochloride (Sigma, St. Louis) was prepared
in normal saline as a stock solution of 128 mg. base
ml.. Stock solution was kept frozen when not in use.
Serial dilutions of the stock solution provided
concentrations of histamine used for the study. Sub-
jects received histamine challenges of 0.1, 2, 4, 8,

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16, 32, 64, and 128 mg./ml. ($9 \times 10^{-4}M$ to $1.15M$; pH=7.0 to 3.7).

  Drugs Used

All drugs were administered single-blindedly.
Cimetidine 300 mg. (Smith, Kline, and French, Philadelphia)
is a highly specific H_2 receptor antagonist (22). It was
selected because of its recent wide use in the treatment
of various illnesses, particularly gastrointestinal
disorders (149). The dosage of the drug provided a
blood level of cimetidine of twice that required to
suppress by 80%, secretion of gastric acid by the
stomach (149).

Chlorpheniramine maleate 8 mg. (USV, Tuckahoe),
an alkylamine derivative, was selected because of its
negligible sedative and anticholinergic effects as well
as its high specificity for H_1 receptors as compared
to other H_1 antihistamines (119).

A lactose placebo dispensed in a pink gelatin cap-
sule (Eli Lilly and Company, Indianapolis) was used.

Medications were administered orally, after an over-
night fast, $1\frac{1}{2}$ to 2 hours prior to histamine challenge, a
time designed to coincide with peak levels of each drug
(127, 130, 149). No side effects of cimetidine, chlorphen-
iramine, or placebo were reported. Since both cimetidine (143,
149) and chlorpheniramine (122) have very short physiological
and pharmacological half-lives, the possibility of

drug-drug interactions was negligible, since the drugs
were administered at least 24 hours apart.

Aerosol Challenge

Histamine was delivered via a Dautrebande D-30
nebulizer (45, 46), driven by 20 pounds per square
inch of compressed air (0.5 ml. liquid nebulized/min.).
Particles of histamine were thus consistently aerosol-
ized to less than 0.5 microns in diameter. This assured
deposition of histamine in the respiratory bronchioles
and lung alveolar spaces.

Subjects wore a noseclip and inhaled the histamine
through a mouthpiece attached to a separate, valved
breathing circuit. Histamine was inhaled for 30 seconds
by tidal breathing. Tidal breathing controlled against
the effects of deep inspiration on pulmonary function
in non-asthmatics (108). Adverse symptoms to inhaled
histamine at highest challenge doses included headache,
cough, flushing of the skin, tachycardia, and chest
tightness. Higher doses of histamine were not given
to subjects after the onset of adverse symptoms.

Pulmonary Function Testing

Histamine induces large airway constriction
through both direct agonism with H_1 receptors, and to
a lesser extent, by vagal mechanisms. Peripheral

airway constriction is almost entirely the result of
direct agonism with H_1 receptors of smooth muscle
(8, 47). Keeping in mind the fact that histamine was
being delivered primarily to small airways, the pulmonary function
tests selected for this experiment were selected for
their sensitivity in measuring small airway constriction.
A particularly sensitive measure of small airways con-
striction is the $MEF_{40}(P)$, the maximum expiratory flow
at 60 percent below vital capacity on a partial expira-
tory curve (20, 105). This test measures flow during
the part of expiration which is independent of expira-
tory effort. The flow at 60 percent of vital capacity
is determined by the static recoil pressure of the lung
and the flow-resistive properties of small airways
(20, 105).

The particular sensitivity of the partial flow-
volume curve may reflect the absence of the effects
of a deep inspiration required for the generation of
maximal expiratory flow volume curves (69, 72, 108).

All subjects were familiarized with the breathing
maneuvers and equipment at the beginning of the study.
Breathing maneuvers were repeated until reproducible
curves could be obtained.

Subjects blew into a cardboard tube attached to
a computerized pneumotachograph-integrator system (148)

which calculated flow and volume. The measurements were recorded on a Gould x-y recorder (slew rate of 40 inches per second). The equipment was calibrated daily.

Subjects expired to residual volume, then inspired to approximately 50-70% of their vital capacity. While wearing a noseclip, the subjects then expired into the cardboard tube as fast as possible to residual volume, thereby generating the partial expiratory flow-volume (PEFV) curve.

Next, subjects inspired to vital capacity and then expired as fast as possible to residual volume, thereby generating the maximal expiratory flow-volume (MEFV) curve. A programmable marker, set to trigger at 1 second, permitted identification of the forced expiratory volume at 1 second (FEV_1). The resultant curves allowed measurement of the forced vital capacity (FVC), peak expiratory flow rate (PEFR), and maximum expiratory flow rate at 60% below vital capacity on the MEFV curve (MEF_{40}) and PEFV curve ($MEF_{40}(P)$). (Figure 1) FEV_1 is a particularly sensitive measure of airway obstruction during a relatively effort-independent portion of the curve.

Protocol

Each day of the experiment, there were controls against factors known to influence responses to histamine (87). Each day, pre-challenge flow-volume maneuvers were performed to establish the baseline for the day. The baseline FEV_1 and $MEF_{40}(P)$ for the subjects on each day was expressed as a percent of the baseline on Day 1. (TABLE II) Histamine challenge was performed at the same time of day to control for diurnal variations in histamine response (49, 95).

Day 1--Histamine Dose Response

On Day 1, the baseline pulmonary function was determined. Prior to each histamine challenge, each subject repeated three flow-volume maneuvers to establish a pre-challenge control value. Starting with doses of 0.1 mg./ml. of histamine, subjects received a histamine challenge as previously described. Inhalation of histamine was followed immediately by pulmonary function testing at 0, 1, 2, 4, 6, and 8 minutes. The averages of the first three and second three blows were calculated.

If a subject did not experience a 20% fall in MEF_{40} (P) as compared to pre-challenge control values, within the first three minutes, then the next successive dose of histamine was administered after a thirty-minute

wait. The wait between successive doses of histamine
challenge was designed to control against any cumula-
tive effect of histamine (87). Pre-challenge controls
expressed as a percentage of the baseline for Day 1
are recorded in TABLE III.

When a subject experienced a 20% decline in
$MEF_{40}(P)$ as compared to pre-challenge control, this
dosage was referred to as the "threshold" dose.
(Figure 2)

Days 2, 3, and 4--Premedication With Antihistamines

On each of Days 2, 3, and 4, subjects were given,
on successive days, an oral dose of H_1 antihistamine
(chlorpheniramine), H_2 antihistamine (cimetidine),
or placebo (lactose), $1\frac{1}{2}$ to 2 hours prior to histamine
challenge.

Baseline pulmonary function was determined at the
beginning of each day. Prior to each histamine challenge,
each subject established a pre-challenge control value
as on Day 1. Pre-challenge controls as a percent of
baseline for Days 2, 3, and 4 are recorded in TABLE IV.

Each subject received a dose of histamine at one
dose below the threshold dose as determined on Day 1
(T-1). Subjects also received a histamine challenge
at threshold dose (T) and one dose above the threshold

(T+1). Each histamine challenge was followed by
pulmonary function testing at 0, 1, 2, 4, 6, and
8 minutes after histamine challenge. The averages of
the first three and second three blows were calculated
for $MEF_{40}(P)$ and FEV_1. The average of the first three
blows was calculated as a percentage of the pre-challenge
control.(TABLE V)(Figures 3 and 4)

Analysis of Data

The means and standard deviations for the pulmonary
function tests were calculated. Using this information,
a t-test for the comparison of two means (58) was used
to compare changes in daily baseline or pre-challenge
control pulmonary function, as well as changes in
pulmonary function following histamine challenge as
compared to pre-challenge control values.

Underlying the comparison of the $MEF_{40}(P)$ values,
is the assumption that the air flow reflected a defined
lung volume. This could be the case only if total lung
capacity remained unchanged during histamine provocation.
Total lung capacity has indeed been shown (20, 141) to
remain constant during induced bronchoconstriction in
asthmatics and normal subjects.

Results

Analysis of the baseline FEV_1 and $MEF_{40}(P)$ for each day (TABLE II) shows that pulmonary function did not significantly vary from day to day.

A clear dose response curve (TABLE VI) could be generated for histamine inhalation. The effects of histamine on pulmonary function for the group became significant at 32 mg./ml. for both the $MEF_{40}(P)$ and FEV_1 curves. The $MEF_{40}(P)$ curve was more significant than the FEV_1 curve at doses of 32 mg./ml. and 64 mg./ ml.. (Figure 2, TABLE VII)

There was a slight cumulative effect of histamine on pre-challenge control levels at 64 mg./ml. of histamine. (TABLE III)

Chlorpheniramine significantly prevented the decline in $MEF_{40}(P)$, at threshold doses of histamine, as compared to Day 1. Pulmonary function values for placebo and cimetidine were not significantly different at threshold as compared to Day 1. (TABLE VIII)

Pretreatment with chlorpheniramine significantly protected against the fall in $MEF_{40}(P)$ as compared to placebo at doses (T) and (T+1). Significant protection of chlorpheniramine against the fall in FEV_1 as compared to placebo was apparent at dose (T+1). There was no significant difference between placebo and cimetidine

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for either $MEF_{40}(P)$ or FEV_1 at any test dose of histamine. (Figures 3 and 4, TABLES V, VIII, and IX)

Discussion

Various conclusions may be drawn from this study. It can be seen from the comparison of baseline pulmonary function from each day that chlorpheniramine, cimetidine, and placebo each had no bronchoconstrictory or broncho-dilatory effects on baseline pulmonary function. Popa (127) studied the effects of oral and intravenous chlor-pheniramine in asthmatics. He found that chlorphenir-amine did improve baseline pulmonary function. However, this study examined the effect of chlorpheniramine on non-asthmatics. The difference in findings may be due to the relative absence of endogenous histamine in non-asthmatics as compared to asthmatics (17) in addition to the decreased sensitivity of non-asthmatics to the little endogenous histamine which may exist (91, 110, 139). Thus, while chlorpheniramine may block H_1 recep-tors in asthmatics which would otherwise be stimulated by histamine, chlorpheniramine blocks receptors in non-asthmatics, which, in the absence of exogenous histamine provocation, would otherwise not be agonized.

Subjects could not be excluded from the study if baseline pulmonary function varied from day to day, as

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this variation might be attributed to the effect of the drug. Exclusion of these persons might therefore skew the results in favor of persons in whom the drugs had no effect.

The finding that baseline pulmonary function did not significantly vary daily added, in retrospect, a useful control to the study. Benson (15) found that the resting state of the airways was an important determinant of bronchial reactivity. Thus, if either chlorpheniramine, cimetidine, or placebo increased the resting bronchomotor tone in the airways, they could have potentiated subsequent bronchoconstriction due to histamine.

It may also be concluded that one dose of 300 mg. of oral cimetidine has no effect on baseline pulmonary function in non-asthmatics. It would be premature to extend this conclusion to the clinical setting of continuous cimetidine use in non-asthmatics. However, this finding suggests the possibility of conducting such a study which would examine pulmonary function in non-asthmatics before, during, and after chronic cimetidine use.

This study confirmed that the methodology used can successfully generate a dose response curve for inhaled histamine provocation. Furthermore, the usefulness of the $MEF_{40}(P)$ pulmonary function test as a

sensitive measure of bronchoconstriction has been demonstrated. The measurement of $MEF_{40}(P)$ is particularly useful in measuring small airways constriction. In addition, the Dautrebande D-30 nebulizer is particularly useful in delivering small histamine particles to the smallest airways. It may thus be concluded that receptors mediating bronchoconstriction, which are stimulated by histamine, exist in the small airways of healthy, human subjects.

A small cumulative effect of histamine was observed to occur on Day 1 as evidenced by pre-challenge pulmonary function, upon reaching a dose of 64 mg./ml., the highest threshold dose of histamine for any subject. However, no cumulative response was apparent on Days 2, 3, and 4. The finding of a cumulative response on Day 1 is surprising. Juniper, et al. (87) found no such response after waiting only five minutes between successive histamine challenges, although they did not challenge anyone with more than 16 mg./ml. of histamine. Perhaps the time required to adequately clear 32 mg./ml. from the respiratory system, as delivered to the airways by the protocol used in this study is greater than the thirty minutes which was permitted before the 64 mg./ml. challenge. The small decline in baseline pulmonary function prior to the

64 mg./ml. challenge might also be attributed to the
general fatigue of the subjects, who spent over two
hours waiting and who previously performed the pulmonary
function maneuveur more than for any other test dose.

Chlorpheniramine had a very significant effect
in preventing induced bronchoconstriction due to
histamine inhalation. Blockade of the H_1 receptors
with the oral medication was significantly greater than
placebo. The difference between chlorpheniramine and
placebo could be detected at both the threshold and
superthreshold doses of histamine. As in the histamine
dose response curve, $MEF_{40}(P)$ was a more sensitive test
than FEV_1 in detecting differences between chlorphenir-
amine and placebo, as well as detecting the protective
effect of chlorpheniramine as compared to the histamine
dose response curve.

Chlorpheniramine did not have a completely protective
effect against the bronchoconstricting effects of hista-
mine. This may have been due, in part, to incomplete
antagonism of all the H_1 receptors in the small airway
by the oral medication. Inhaled H_1 receptor antagonists,
such as clemastine (113) have apparently been more
successful than oral medications in providing H_1
antagonism in asthmatics. In fact, clemastine (113)
causes bronchodilation in asthmatics, perhaps by leaving

endogenous histamine free to stimulate bronchodilating H_2 receptors.

Chlorpheniramine is a competitive antagonist of H_1 receptors. Thus, high concentrations of histamine, such as those which may have been by the T+1 dose, could displace the antagonist from the receptor.

A small component on the bronchoconstriction due to histamine may have been the result of vagally-mediated, reflex bronchoconstriction (52). Histamine particles, particularly those with a moderately acidic pH among the higher doses, may have stimulated irritant receptors, particularly in the larger airways (47) to cause bronchoconstriction which can not be prevented by H_1 antagonism. In this study, parasympatholytic agents, such as atropine, may have provided additional protection.

Unlike inhaled clemastine (113), chlorpheniramine did not yield any bronchodilation either at baseline or upon histamine challenge. This result does not support the presence of H_2 receptors in the normal human lung which mediate bronchodilation.

The absence of an H_2 receptor-mediated broncho-dilation at baseline may be explained as due to either the absolute deficiency of H_2 receptors, or more likely, the absence of endogenous histamine in non-asthmatics which could bind with H_2 receptors. The fact that histamine challenge did not yield bronchodilation

during H_1 receptor blockade, may be due to the absolute lack of H_2 receptors. It may also be possible, that with the oral medication, even at subthreshold levels of histamine, there were still an adequate number of unbound H_1 receptors agonized by histamine to counter any effect of H_2 agonism. Thus, a net bronchoconstriction would be observed.

Further studies in the non-asthmatic might explore the possible bronchodilatory effects of an inhaled antihistamine on both baseline pulmonary function and response to histamine challenge. Smaller doses of histamine than those which were used in this study might simulate endogenous levels of histamine in the asthmatic and agonize any possibly existent H_2 receptors without displacing H_1 antagonists and causing a net H_1 receptor-mediated bronchoconstriction. Pretreatment with an H_2 antagonist in addition to an H_1 antagonist on the same day might remove H_2-mediated bronchodilation, resulting in a significantly larger net bronchoconstriction than in pretreatment with only an H_1 antagonist.

No placebo effect was observed. Placebos have been reported to influence the amount of bronchoconstriction due to histamine provocation (131). However, the placebo has to be presented to the subject as a substance which has a bronchoconstricting or broncho-

dilating effect. Subjects in this study were not informed as to what the effects of the drugs might be with regard to subsequent histamine challenge.

Cimetidine was no different than placebo with regard to pulmonary finction observed upon histamine challenge. Blockade of H_2 receptors neither protected against or potentiated bronchoconstriction. If H_2 receptors, which mediated bronchoconstriction, existed in the lung, then cimetidine would be expected to have a protective effect, as was the case with chlorpheniramine.

If H_2 receptors which mediate bronchorelaxation were successfully anatagonized by cimetidine, then subsequent histamine challenge would stimulate only H_1 receptors. This would promote an even greater bronchospasm than that observed with placebo, in which case, both H_1 and H_2 receptors are stimulated.

The H_2 receptor has been implicated, through cyclic AMP mechanisms, to have a variety of effects which would inhibit the effects of histamine challenge. These effects include H_2 receptor-mediated inhibition of further histamine release (94), inhibition of histamine production (104, 125), promotion of histamine metabolism (67, 107, 125), promotion of histamine clearance from the circulation (34, 133), and promotion

of histamine uptake by mast cells (34, 68, 107).
Blockade of these effects, particularly the effects
on metabolism, uptake, and clearance, by cimetidine
would also potentiate histamine-induced bronchospasm.
However, cimetidine was not successful in potentiating
histamine-induced bronchospasm.

These findings are consistent with the preliminary
reports of Eiser (59) and Maconochie (99) who also
found no bronchoconstrictory effects of cimetidine.
The most likely conclusion to be drawn from the evi-
dence is that H_2 receptors do not exist to any signi-
ficant degree in non-asthmatic airways. If H_2 receptors
are present in the lung, the effects of H_2 agonism are
negligible compared to the bronchoconstrictory effects
of H_1 agonism.

If there are few H_2 receptors in the lung, it is
possible that at the doses of histamine used for this
experiment, the histamine overcame the competitive
antagonism by cimetidine for H_2 receptor sites in the
lung. Thus, cimetidine might have been displaced from
the opportunity to potentiate bronchospasm. At lower
levels of histamine agonism, H_2 blockade might have
potentiated bronchospasm. While it is difficult to
measure bronchospasm in non-asthmatics at low levels
of histamine challenge, asthmatics are much more

sensitive to low levels of histamine (91, 110, 139).
The finding that cimetidine promotes bronchospasm in
asthmatics (71, 109) may be due to the fact that
cimetidine is not displaced by the low levels of
histamine used in the histamine challenges. In addi-
tion, acting through mechanisms which potentiate the
presence and release of histamine (34, 67, 68, 94,
104, 125, 133) small levels of additional histamine
which would not provoke bronchospasm in the non-
asthmatic might potentiate bronchospasm in the sensi-
tive asthmatic.

It is possible, as has been pointed out by
Macanochie, et al. (99) that the cimetidine used in
this study did not achieve adequate levels in the
lung. Cimetidine levels in human lung following oral
administration have not been studied. The effects of
cimetidine on histamine metabolism, uptake and clearance
have also not been investigated. However, the dosage
of cimetidine used in this study provided a blood
level of twice that required to inhibit gastric acid
secretion in the stomach by 80%.

Perhaps future studies could investigate the effects
of an inhaled H_2 agonist, such as 4-methylhistamine or
dimaprit on pulmonary function. Future studies could
also investigate the effects of an inhaled H_2 antagonist,

in hopes of achieving better penetration in the lung
than with oral administration.

A possible reason for the findings of this investi-
gation may be the failure of cimetidine to adequately
antagonize the histamine receptors in the lung which
mediate bronchodilation. A previous study (66) demon-
strated that cimetidine did not potentiate the depressor
actions of histamine in tissue in which burimamide and
metiamide, which belong to a different class of H_2
antagonists, potentiated the depressor effects on carotid
blood pressure. Similarly, a subclass of H_2 receptor
not antagonized by cimetidine, might be present in the
human lung.

Several other studies have demonstrated the
presence of histamine receptors in the airways of the
rat, ferret (40), cat (35), horse (37, 39), and rabbit
(36, 70) which mediate bronchodilation and can be
agonized by 4-methylhistamine (40), yet which cannot
be antagonized by conventional H_2 antihistamines.
Perhaps such a class of H_3 receptors may exist in human
lung.

Histamine was aerosolized so as to reach the smallest
airways (45, 46). The $MEF_{40}(P)$ pulmonary function test
is particularly sensitive to small airways constriction
(20, 105). If H_2 receptors in the human lung exist

predominantly in the large airways, the protocol of this investigation may not have been sensitive enough to document any effects of H_2 blockade. Regional differences in the distribution of H_2 receptors has been demonstrated in several animal species (14, 64, 156). For example, H_2 histamine receptors are more prominent in the central airways than the peripheral airways in the guinea pig (156).

The regional differences in histamine receptor distribution may correlate with the physiologic importance of maintaining the patency of the large airways. An alternative study could examine the effects of larger aerosolized histamine particles on pulmonary flow resistance, a sensitive measure of central resistance (98), in order to more closely investigate the presence of H_2 receptors in large airways. Atropine would have to be used in this proposed protocol, because of the relative importance of irritant receptors in the large airways.

In conclusion, the experimental evidence presented in this investigation confirms the presence of H_1 receptors in the airways of healthy human subjects. These receptors appear to mediate bronchoconstriction and can be successfully antagonized by chlorpheniramine.

The role of H_2 receptors in healthy human airways, as either bronchodilators acting directly on smooth muscle or as inhibitors of the presence of histamine, appears to be negligible under the conditions of the protocol used in this study. Cimetidine did not potentiate histamine-induced bronchospasm. The role of H_2 receptors in asthmatic bronchoconstriction and bronchodilation remains a possibility for future investigation.

MAXIMAL AND PARTIAL EXPIRATORY FLOW-VOLUME CURVES
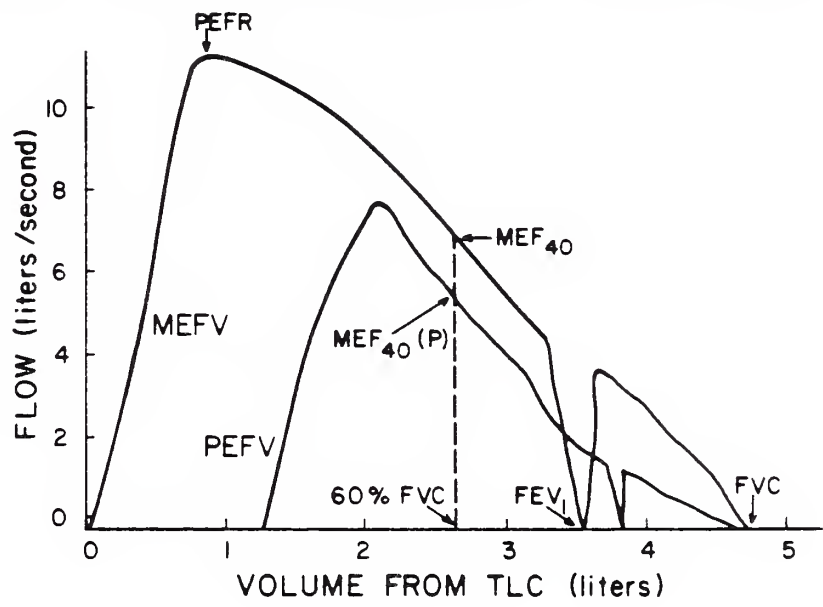
Figure 1

HISTAMINE DOSE RESPONSE AS CHARACTERIZED
BY TWO MEASUREMENTS OF PULMONARY FUNCTION
(FEV$_1$ and MEF 40% (P))
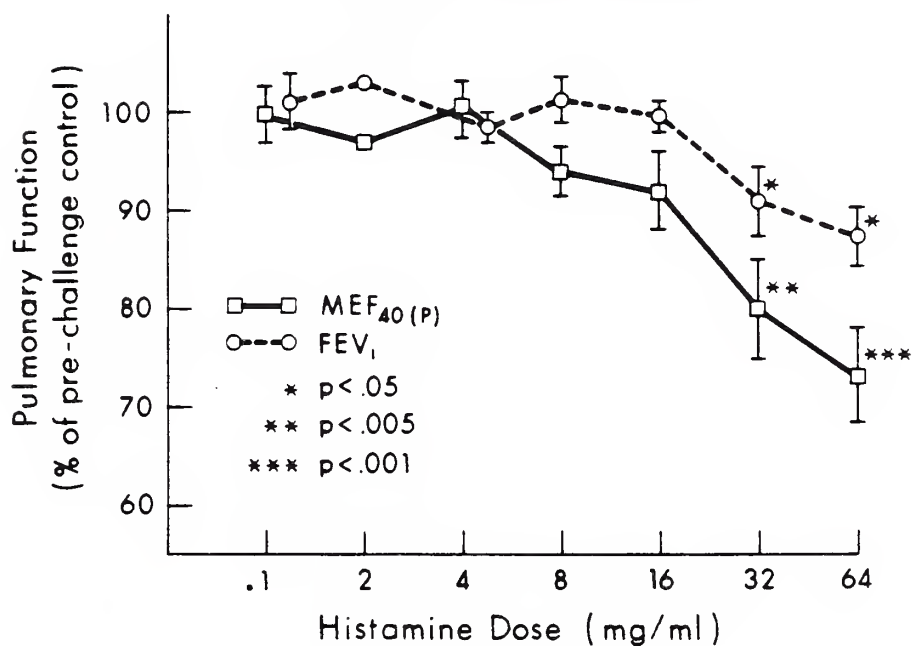
Figure 2

THE EFFECT OF ANTIHISTAMINES
ON HISTAMINE CHALLENGE AS MEASURED
BY MEF 40% (P)
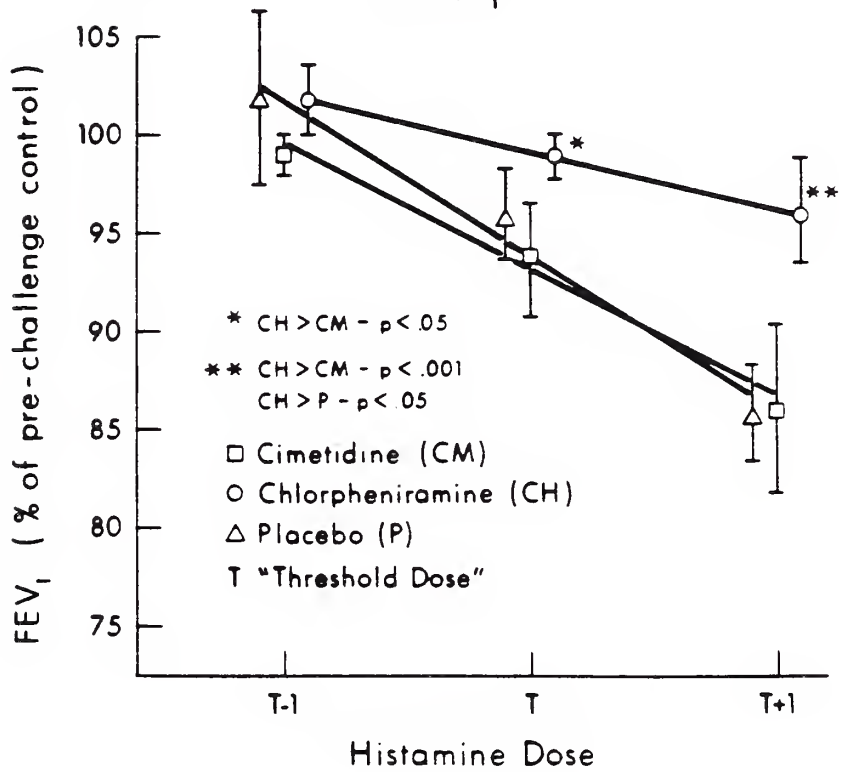
Figure 3

Figure 4

TABLE I

ANTHROPOMORPHIC DATA

	FVC	FEV_1	$FEV_1\%$	PEFR	$MEF_{50}\%$	Age
Mean	4.41	3.68	83.9	8.21	4.50	26.3
Standard deviation	0.82	0.69	9.3	1.62	2.10	5.1
Percent of expected values	89%	94%		93%	102%	

TABLE II

BASELINES AT BEGINNING OF DAY
AS PERCENTAGE OF BASELINE ON DAY 1

DAY	1	CM	CH	P
FEV_1	100	99.6	99.8	99.0
$MEF_{40}(P)$	100	98.1	104.6	109.9

CM=Cimetidine
CT=Chlorpheniramine
P =Placebo

TABLE III

AVERAGE HISTAMINE DOSE-RESPONSE
PRE-CHALLENGE CONTROLS AS
PERCENT OF INITIAL BASELINE

DOSE	.1	2	4	8	16	32	64
FEV_1	100	100	98.9	98.1	98.6	99.0	100.7
$MEF_{40}(P)$	100	110	94.1	102.4	98.1	93.9	89.0*

*p= .031

TABLE IV

PRE-CHALLENGE CONTROLS ON DAYS 2, 3, AND 4
AS PERCENT OF INITIAL BASELINE

CIMETIDINE

DOSE	T-1	T	T+1
FEV_1	100	98.45	98.6
$MEF_{40}(P)$	100	101.4	101.9

****

CHLORPHENIRAMINE

DOSE	T-1	T	T+1
FEV_1	100	100.36	100.8
$MEF_{40}(P)$	100	102.3	98.4

****

PLACEBO

DOSE	T-1	T	T+1
FEV_1	100	99.5	102.0
$MEF_{40}(P)$	100	97.0	95.4

T=Threshold Dose

TABLE V

THE EFFECT OF PRETREATMENT WITH ANTIHISTAMINE
UPON SUBSEQUENT RESPONSE TO HISTAMINE

(Means $\pm$ Standard Error of the Mean)

FEV_1

DOSE	T-1	T	T+1
CIMETIDINE	99.0 ± 1.13	93.64 ± 2.79	86.0 ± 4.30
CHLORPHENIRAMINE	101.72 ± 1.71	98.91 ± 1.23	96.09 ± 2.60
PLACEBO	101.88 ± 4.42	95.75 ± 2.54	85.88 ± 2.49

$MEF_{40}(P)$

DOSE	T-1	T	T+1
CIMETIDINE	96.0 ± 4.26	79.64 ± 3.70	57.80 ± 7.39
CHLORPHENIRAMINE	99.0 ± 4.17	91.45 ± 2.54	82.18 ± 5.58
PLACEBO	91.88 ± 3.66	78.13 ± 5.72	57.88 ± 5.15

TABLE VI

HISTAMINE DOSE RESPONSE
AS PERCENTAGE OF PRE-CHALLENGE CONTROL

(Means ± Standard Error of the Mean)

DOSE	.1	2	4	8	16	32	64
FEV_1	101.1 ±2.65	103 ±0	98.63 ±1.39	101.6 ±2.01	99.45 ±0.73	91.0 ±3.59	87.57 ±2.98
$MEF_{40}(P)$	99.90 ±2.68	97 ±0	100.5 ±3.17	94.2 ±2.46	92.0 ±3.90	79.72 ±4.92	73.29 ±4.53

TABLE VII

RESPONSE TO INHALED HISTAMINE
AS COMPARED TO RESPONSE TO
0.1 MG./ML. OF INHALED HISTAMINE

DOSE	4	8	16	32	64
FEV_1	NS	NS	NS	$p<.05$	$p<.05$
$MEF_{40}(P)$	NS	NS	$p=.10$	$p<.005$	$p<.001$

NS=No significance

TABLE VIII

COMPARISON OF THE PROTECTIVE EFFECT
OF CHLORPHENIRAMINE, CIMETIDINE, AND PLACEBO
UPON SUBSEQUENT RESPONSE TO INHALED HISTAMINE
AS MEASURED BY $MEF_{40}(P)$

	DAY 1	CHLORPHENIRAMINE(CT)	CIMETIDINE(CM)	PLACEBO(P)
DAY 1				
T-1	*****	NS	NS	NS
T	*****	p<.0025	NS	NS
T+1	*****	*	*	*
CT				
T-1	NS	*****	NS	NS
T	p<.0025	*****	p<.01	p<.05
T+1	*	*****	p<.0005	p<.005
CM				
T-1	NS	NS	*****	NS
T	NS	p<.01	*****	NS
T+1	*	p<.0005	*****	NS
P				
T-1	NS	NS	NS	*****
T	NS	p<.05	NS	*****
T+1	*	p<.005	NS	*****

*Insufficient data to make comparison

NS=No significance

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TABLE IX

COMPARISON OF THE PROTECTIVE EFFECT
OF CHLORPHENIRAMINE, CIMETIDINE, AND PLACEBO
UPON SUBSEQUENT RESPONSE TO INHALED HISTAMINE
AS MEASURED BY FEV_1

	DAY 1	CHLORPHENIRAMINE(CT)	CIMETIDINE(CM)	PLACEBO(P)
DAY 1				
T-1	*****	p=.07	NS	NS
T	*****	p=.14	NS	NS
T+1	*****	*	*	*
CT				
T-1	p=.07	*****	NS	NS
T	p=.14	*****	p<.05	NS
T+1	*	*****	p<.001	p<.05
CM				
T-1	NS	NS	*****	NS
T	NS	p<.05	*****	NS
T+1	*	p<.001	*****	NS
P				
T-1	NS	NS	NS	*****
T	NS	NS	p<.05	*****
T+1	*	p<.05	p<.001	*****

*Insufficient data to make comparison
NS=No significance

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